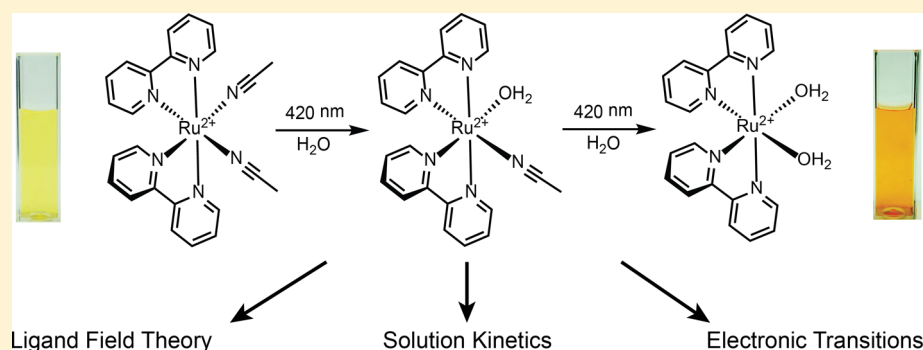


Kinetics and Photochemistry of Ruthenium Bisbipyridine Diacetonitrile Complexes: An Interdisciplinary Inorganic and Physical Chemistry Laboratory Exercise

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S Supporting Information



ABSTRACT: The study of ruthenium polypyridyl complexes can be widely applied across disciplines in the undergraduate curriculum. Ruthenium photochemistry has advanced many fields including dye-sensitized solar cells, photoredox catalysis, light-driven water oxidation, and biological electron transfer. Equally promising are ruthenium polypyridyl complexes that provide a sterically bulky, photolabile moiety for transiently “caging” biologically active molecules. Photouncaging involves the use of visible (1-photon) or near-IR (2-photon) light to break one or more bonds between ruthenium and coordinated ligand(s), which can occur on short time scales and in high quantum yields. In this work we demonstrate the use of a model “caged” acetonitrile complex, $\text{Ru}(2,2'\text{-bipyridine})_2(\text{acetonitrile})_2$, or RuMeCN in an advanced synthesis and physical chemistry laboratory. Students made RuMeCN in an advanced synthesis laboratory course and performed UV–vis spectroscopy and electrochemistry. The following semester students investigated RuMeCN photolysis kinetics in a physical chemistry laboratory. These two exercises may also be combined to create a 2-week module in an advanced undergraduate laboratory course.

KEYWORDS: Upper-Division Undergraduate, Inorganic Chemistry, Interdisciplinary/Multidisciplinary, Laboratory Instruction, Physical Chemistry, Hands-On Learning/Manipulatives, Aqueous Solution Chemistry, Chromatography, Coordination Compounds, Crystal Field/Ligand Field Theory

Chemical education has a need for interdisciplinary laboratory courses that expose students to exciting new applications of chemistry while preparing them to solve real-world problems that cross multiple scientific disciplines.^{1–3} As a result, there has been an increase in the publication of new, strongly interdisciplinary laboratory experiments that span the classical divisions of chemistry; from organic/inorganic^{4,5} to physical/inorganic² and biological/organic⁶ examples, these experiments have begun to transform the undergraduate chemical laboratory curriculum.

Ruthenium compounds of the general form $\text{Ru}(\text{polypyridyl})_2(\text{L})_2$, where L is any monodentate ligand coordinated via N or S, experience photoactivated ligand (L) exchange with coordinating solvents (such as water) due to a thermally accessible triplet metal-centered state (Figure 1).^{7,8} If the ligands (L) are biologically active, this property is useful for photoreleasing a drug with high spatial and temporal control.⁹ Even if the ligands are chemically inert, the ruthenium product $[\text{Ru}(\text{polypyridyl})_2-$

$(\text{H}_2\text{O})_2]^{2+}$ itself is biologically active: by binding irreversibly to DNA bases in a manner similar to cisplatin, it can induce cell apoptosis, which provides another opportunity to spatially target drug delivery.¹⁰

Photoactive ruthenium complexes recently described in the literature have used chelating polypyridyl ligands with extended pi systems to red-shift the absorbance of the complex and allow for photolysis within living tissue.^{10,11} However, to facilitate synthesis and characterization for an undergraduate laboratory, we worked with a model compound RuMeCN , which is readily synthesized from commercially available $\text{Ru}(\text{bpy})_2\text{Cl}_2$ ^{12,13} (where bpy is 2,2'-bipyridine) and acetonitrile and is stable in red light, which makes it practical for student handling in a large laboratory setting. Upon continuous irradiation of RuMeCN

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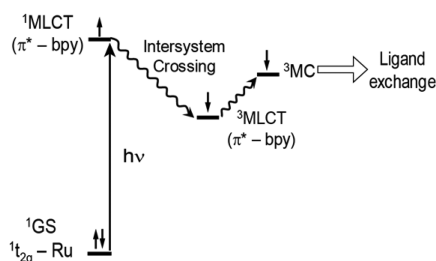


Figure 1. A typical Jablonski diagram that depicts the electronic transitions responsible for light-mediated ligand loss observed in many Ru(2,2'-bipyridine)₂(L)₂ compounds. An electron is excited from the ground state (¹GS) on the metal center into a metal-to-ligand charge transfer state (¹MLCT). It undergoes rapid intersystem crossing into a triplet MLCT state, from which it can populate the thermally accessible triplet metal-centered state, ³MC, responsible for ligand exchange.

under blue light, both CH₃CN ligands are exchanged sequentially, which provides a “textbook example” of A → B → C kinetics. Multistep kinetic processes are common in nature but can be difficult to monitor spectroscopically. The RuMeCN photolysis reaction provides a valuable opportunity to collect and analyze data for a model two-step kinetic process.

In addition to its symmetrical octahedral structure and beautiful yellow color, RuMeCN is distinctive for the pedagogical opportunities it provides. Synthesis of this molecule teaches students basic Schlenk line techniques on a system that is less sensitive to oxygen and water than many other inorganic syntheses. RuMeCN is also reversibly and cleanly oxidized and reduced, and cyclic voltammetry is useful for characterizing the electronic transition that gives rise to ligand exchange.¹⁴ Students observe the characteristic MLCT band of RuMeCN and photolysis products by UV–vis spectroscopy, which gives an opportunity to discuss electronic transitions and ligand-field theory in detail. Lastly, quantitative analysis of the two-step ligand exchange process, coupled with equation derivation and data fitting, complements the physical chemistry lab course. In completing these laboratory exercises with RuMeCN, students gain the requisite skills to design, synthesize, and characterize a wide variety of inorganic compounds, especially novel Ru-caged compounds, which could be developed as a separate laboratory exercise.

Other experiments have been published recently that use ruthenium chemistry to cross disciplines in a similar manner. For example, this laboratory exercise complements previous studies of these Ru complexes and their photoactivity by DFT and computational modeling.¹⁵ Similarly, because ruthenium coordination compounds have been widely applied in the biological sciences, another interdisciplinary experiment includes the use of ruthenium “piano-stool” complexes in DNA binding and cleavage.¹⁶ In our case, we applied the photochemical reaction of Ru(bpy)₂(CH₃CN)₂ to advanced laboratory courses in both inorganic synthesis and experimental physical chemistry. Because of course scheduling, many students performed the synthesis laboratory one semester before the physical chemistry laboratory; thus, these very complementary experiments were designed to stand alone.

The pedagogical goals for the two experiments are described here as follows: for advanced synthesis, students will (1) perform a reaction on a Schlenk line and purify product by column chromatography, (2) characterize the compound by NMR and cyclic voltammetry, (3) determine a molar

absorptivity and describe the electronic transition observed, and (4) demonstrate the application of ligand field theory in an experimental system. For physical chemistry, the goals were to (1) provide students with an experimental system with complicated kinetics mechanism, (2) fit data from experiments to formulas derived from basic equations, and (3) discuss the physical meaning of a quantum yield.

■ INORGANIC SYNTHESIS LABORATORY (8 H TOTAL, TWO 4-H SECTIONS)

Synthesis and Characterization of [Ru(bpy)₂(CH₃CN)₂]Cl₂ (RuMeCN)

Laboratory sections consisted of 21 students working in lab groups no larger than three. The course was supervised by an instructor and two graduate student TAs. The first 4-h lab period was used to synthesize and purify RuMeCN, and a second 4-h lab period was used for characterization.

Synthesis was performed on a 100 mg (0.192 mmol) scale of starting material Ru(bpy)₂Cl₂ (Supporting Information, Section 1B), which gave an average yield of 60%, or ~65 mg of product per group. RuMeCN was characterized by ¹H NMR in CD₃CN. This same sample was then used for electrochemical characterization in CH₃CN with TBAPF₆ as the electrolyte (Supporting Information, Section 1C). UV–vis spectroscopy was performed in water with concentrations ranging from 0.03–0.2 mM. Molar absorptivity was determined by a Beer–Lambert Law plot, and the ligand exchange was observed by UV–vis spectroscopy upon irradiation with several different light sources including white LED flashlights and cell phone flashlights.

■ PHYSICAL CHEMISTRY LABORATORY (4-H SECTION)

Kinetics Studies

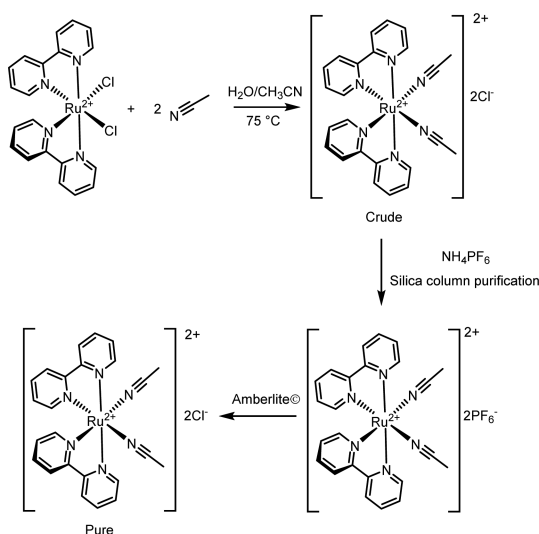
Laboratory sections for physical chemistry consisted of 26 students supervised by an instructor and two graduate student TAs. Students worked in pairs to collect UV–vis absorbance spectra showing the spectral shift under visible light irradiation, and to determine how long the kinetics trials would take, then collected three kinetics traces observing the absorbance at 450 nm under continuous irradiation.

[Ru(bpy)₂(CH₃CN)₂]Cl₂ was synthesized by a lab technician, and students were given 7 mg aliquots for their use. Stock (1 mM) solutions of RuMeCN were made, from which students made aliquots to generate photolysis solutions of around 0.03–0.1 mM. Blue presentation laser pointers (max wavelength 405 nm) were used as light sources. The power of the light source was measured before each kinetics trial using a ThorLabs P-100 power meter.

■ HAZARDS

Synthesis should be performed in the hood under inert atmosphere. Gloves, lab coats, and goggles should be worn at all times when synthesizing and purifying the compound. Gloves and goggles should be worn when characterizing and performing kinetics studies. Dry silica gel is an inhalation hazard and should be handled in the hood or with face masks. Dichloromethane is an eye and skin irritant and potentially carcinogenic. Ru(bpy)₂(CH₃CN)₂ is not considered extremely toxic but should be handled with care.

Scheme 1. Synthesis and Subsequent Metathesis Reactions To Form the Final Product RuMeCN



RESULTS

Advanced Synthesis Lab: Complex Synthesis

Synthesis on the 100 mg scale provided most groups with sufficient material for all experiments in the advanced synthesis Lab. The synthesis was performed in the dark by covering round-bottom flasks with foil and turning off the lights. We found it helpful to use desk lamps fitted with red incandescent bulbs to safely illuminate the laboratory during the experiment. The heating step was performed under inert atmosphere (N_2 or Ar) to prevent the formation of reactive oxygen species at the ruthenium, which can lead to side products or degradation during the synthesis.

Once the solution turned orange (as observed by briefly exposing to dim white light), the counterions were exchanged by adding solid NH_4PF_6 and extracting the resultant precipitate with dichloromethane. $[RuMeCN](PF_6)_2$ is soluble in less polar organic solvents (useful for column chromatography), while $[RuMeCN]Cl_2$ is soluble in water and more polar solvents such as methanol and acetonitrile. Students checked the suggested solvent ratio and the purity of the reaction by TLC before the column. Once pure, a metathesis was performed again by passing $[RuMeCN](PF_6)_2$ dissolved in methanol through an anion exchange resin (Amberlite). This was then divided into two vials and dried down separately by rotary evaporation.

Advanced Synthesis Laboratory: Characterization

One vial of $[RuMeCN]Cl_2$ was used to collect 1H NMR spectrum and cyclic voltammogram (Figure 2, Figure S2), as the sample cannot be reused after the electrochemistry experiment. The NMR spectrum showed the purity of the sample, and students were able to calculate the reduction potential, $E_{1/2}$, for the Ru(III) \rightarrow Ru(II) redox couple. It has been suggested in the literature that this value trends with the quantum yield of ligand release.¹⁴

The contents of the other vial, which must be completely dry, was dissolved in water to make a stock solution of approximately 1–5 mM. The molar absorptivity at 420 nm, ϵ_{420} , was determined by generating a standard curve and fitting to Beer's Law as well as a series of spectra showing the spectral shift under constant irradiation (Figure 3). For this lab, many different light sources were used, which varied from a simple

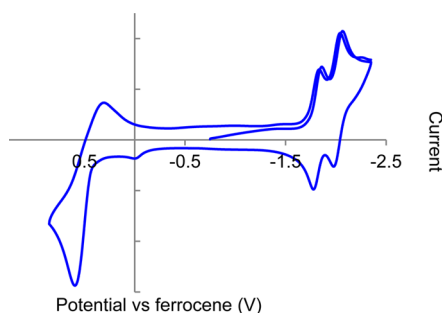


Figure 2. Cyclic voltammogram of RuMeCN in acetonitrile. The two peaks at -1.75 V and -2.00 V correspond to sequential bpy/bpy⁻ reduction for each bpy ligand, and the major redox event at approximately $+0.5$ V corresponds to the Ru(III)/Ru(II) couple.

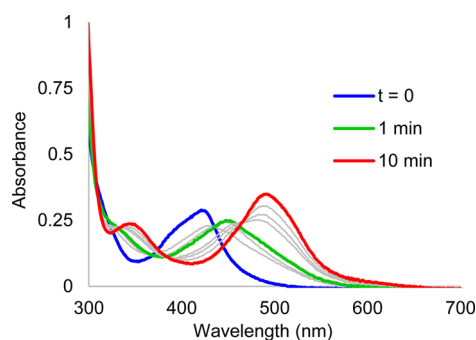


Figure 3. UV-vis spectra of RuMeCN dissolved in water showing the shift in absorbance under continuous irradiation with a bright light source (1–5 mW).

white LED flashlight to cell phone flashlights. It was found that a less powerful light source gives the clearest shift, though this takes longer for the reaction to go to completion (10–20 min). Student results for the molar absorptivity, as well as their average overall synthetic yield, are presented in Table 1.

Table 1. Percent Yield, $E_{1/2}$, and Molar Absorptivity Values Determined for RuMeCN

Values	% Yield	$E_{1/2}^a$ (V vs Fc)	ϵ_{420} ($M^{-1} cm^{-1}$)
Measured	80–96	1.0 ^c	7300
Students ^b	60 \pm 34	0.8 \pm 0.2	5000 \pm 3000

^aIn acetonitrile, with TBAPF₆ as the electrolyte. ^bAverage of 12 groups. ^cPinnick et al.¹²

Physical Chemistry Laboratory: Kinetics

To visualize the change in absorbance expected at 450 nm (where the intermediate species B absorbs most strongly), students again collected a series of UV-vis spectra (similar to Figure 3, using a 405 nm laser pointer, ~ 5 mW). Once students had identified a suitable time scale for their kinetics experiments, they set up the spectrometer for constant irradiation while stirring. Blue (405 nm) laser pointers were suspended over the cuvette, which was placed in the spectrometer on a stir plate, and equipped with a stir bar. The students collected at least three good kinetics traces to reduce experimental error.

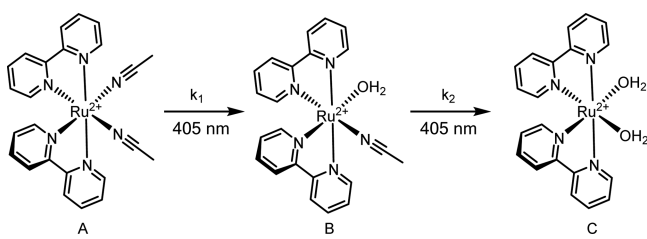
The data were then fit to an equation of the form

$$Abs = A_1 e^{-t/\tau_1} + A_2 e^{-t/\tau_2} + y_0 \quad (1)$$

which was derived from the integrated rate laws

$$[A] = [A]_i e^{-k_i t} \quad (2)$$

Scheme 2. RuMeCN Photolysis, Investigating Observed First (k_1) and Second (k_2) Ligand Exchange Rates



$$[B] = \frac{k_1}{k_2 - k_1} (e^{-k_1 t} - e^{-k_2 t}) [A]_i \quad (3)$$

$$[C] = \left(1 + \frac{k_1 e^{-k_1 t} - k_2 e^{-k_2 t}}{k_2 - k_1} \right) [A]_i \quad (4)$$

(full derivation in [Supporting Information](#), Section 1C). Excel Solver was used to fit the data, given a set of sample values for the constants A_1 , τ_1 , A_2 , and τ_2 . Most students found an acceptable fit, based on the sum of the differences squared between experimental and calculated values, and a visual plot of their data ([Figure 4](#)). Various troubleshooting tips are discussed in the [Supporting Information](#).

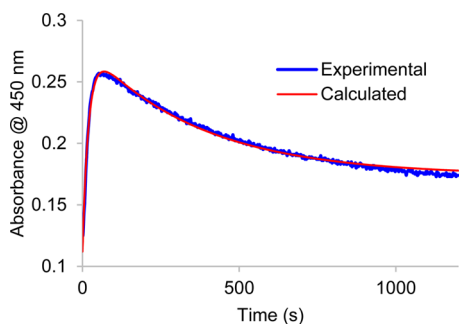


Figure 4. Kinetics trace collected by students of the absorbance measured at 450 nm under constant irradiation with a blue laser pointer (~ 5 mW), plotted against the calculated values determined by Solver.

Physical Chemistry Laboratory: Quantum Yield

The quantum yield calculation required measurement of the power of the light source using a conventional power meter (Thorlabs PM100A). The power of the light source was determined and recorded prior to every kinetics experiment, as the laser pointer power fluctuated somewhat over time.

By using the observed rate constant for the first photolysis step, k_1 , as determined by the data fit, the power, and wavelength of the light source, the quantum yield for the first ligand photorelease, ϕ_{pr} , was calculated:

$$\phi_{pr} = \frac{dA \text{ (moles)}}{d\text{photon (moles)}} = \frac{k_1 [A]_i V_{\text{sample}}}{\frac{P}{E_{ph} N_A}} \quad (5)$$

where $[A]_i$ is the molar concentration of RuMeCN in the cuvette, V_{sample} is the volume (in L) of solution in the cuvette, P is the measured laser power (J/s), E_{ph} is the energy of the photons (hc/λ , in J), and N_A is Avogadro's number (mol^{-1}).

DISCUSSION

Upon completion of the advanced synthesis laboratory, students were required to compile their data and present it in a basic

laboratory report with a paragraph discussing their findings and potential applications for this compound. Because of the report requirements in the physical chemistry laboratory, students in that class were required to submit a written, full-length laboratory report, including a discussion of their results and the viability of this compound for use in photodynamic therapy, especially regarding photolytic reactivity (related to the magnitude of the quantum yield). Most students found both laboratories to be a valuable learning experience and engaging on many levels. Students in the physical chemistry laboratory were asked to rate their levels of understanding concerning certain topics before and after the experiment ([Figure 5](#)), and their answers

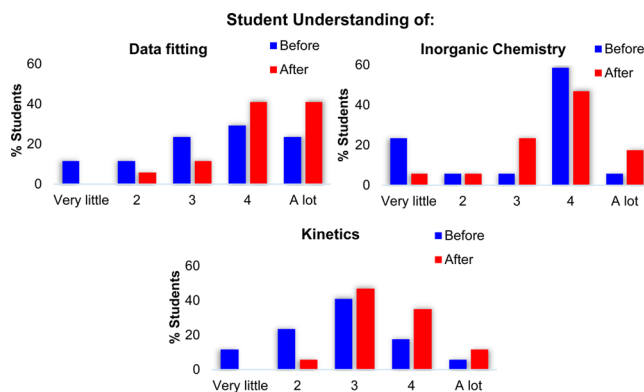


Figure 5. Student feedback after the experiment. Students were asked to compare their knowledge on the subjects of data fitting, ligand field theory and inorganic chemistry, and higher order kinetics before and after the experiment performed in the physical chemistry laboratory (26 students).

showed a positive trend in postlab comprehension levels, with the greatest improvement in knowledge of data fitting. The quality of student data was generally good (see [Tables 1](#) and [2](#)), with several outliers especially in the calculation of molar

Table 2. Observed Rate Constants and Quantum Yield Values

Values	k_1 (s^{-1})	k_2 (s^{-1})	ϕ_{pr}^a
Measured	0.33	0.006	0.51
Students ^b	0.1 ± 0.1	0.008 ± 0.004	0.4 ± 0.3

^aQuantum yield of photorelease. ^bAverage of 13 groups.

absorptivity and the observed rate constants. This was generally due to errors in solution concentration, for example, if the initial mass was incorrect or students failed to dissolve 100% of their sample. The differences in students' experimental values for both observed rate constants, k_1 and k_2 , can be explained by differences in the number of photons delivered to the sample in the different groups (with variability in laser pointer power, illumination path, and stirring efficiency being three important variables).

Further investigations with this system could include performing COSY 2D NMR to assign the peaks in the aromatic region of the proton NMR. Students also suggested exploring other ligand exchange systems and comparing spectroscopic results of the light reaction performed in water or in another coordinating solvent or with different N or S ligands.

■ ASSOCIATED CONTENT

📄 Supporting Information

The Supporting Information is available on the ACS Publications website at DOI: 10.1021/acs.jchemed.6b00173.

Detailed synthetic procedures; laboratory setup; troubleshooting and equation derivation; sample student handouts (PDF, DOCX)

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Notes

The authors declare no competing financial interest.

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